BEHAVIORAL RESPONSES OF TICKS TO MOTION AND REPELLENTS OR ATTRACTANTS

By

JIMMY L. YOUNG

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Oklahoma State University

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BEHAVIORAL RESPONSES OF TICKS TO MOTION AND
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Thesis Approved:

D E Howell
Thesis Adviser

R R Walton

Robert D Morrison

D D Durham
Dean of the Graduate College

PREFACE

A method of evaluating and screening chemicals, repellents, or attractants under laboratory conditions was selected as a thesis problem by the author. The experimentation was designed to determine the behavioral patterns of ticks when placed on a turntable and when the ticks were also subjected to chemically treated filter papers.

The author wishes to express his appreciation to his major adviser, Dr. D. E. Howell, for his guidance throughout the study and in preparation of this paper. Sincere thanks are expressed to Dr. Robert D. Morrison, Professor of Mathematics and Statistics, for his thoughtful guidance and assistance in the statistical designs of the experiment and for his constructive criticism of the thesis manuscript. Also, sincere thanks are expressed to Dr. R. R. Walton, Professor of Entomology, for his valued criticism of the thesis manuscript. Indebtedness is expressed to Berry D. Segal, graduate student in Mechanical Engineering, Dr. Donald E. Boyd, Assistant Professor of Civil Engineering, Dr. A. H. Soni, Assistant Professor of Mechanical Engineering, and Dr. Henry R. Sebesta, Assistant Professor of Mechanical Engineering for help in explaining the mechanics and the effects of coriolis acceleration. Sincere thanks are expressed to Henry Magalit, graduate student in Mathematics and Statistics, for his assistance in the data processing. A special note of thanks is extended to Dr. R. O. Drummond, USDA Laboratory, Kerrville, Texas, for supplying nymphal and adult ticks. A gratitude of thanks is expressed to Mr. Robert L. Heister, Stillwater, Oklahoma, and Mr. Joe Dodrill, McCloud, Oklahoma, for their assistance in collecting replete female ticks. My sincere acknowledgment is made to the United States Army for making this study possible. Gratefulness is also extended to my wife, Mary Pat, for her patience and encouragement.

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INTRODUCTION

This research project was centered around the need for a method to effectively screen tick repellents and/or attractants under laboratory conditions.

In an attempt to do this a turntable was used to equalize the effects of temperature, relative humidity, and light.

A search through the literature revealed that very little work had been done on the behavioral responses of ticks when placed on a turntable, much less their response to a turntable plus chemicals.

Many uniformity trials were conducted to determine the behavioral responses of ticks when placed in a container located near the edge of the turntable. A particular behavioral pattern resulted.

With the above points in mind this research program was designed to account for the various movement pattern of the ticks. Five repellents, each at three concentrations, were tested with *Dermacenter albipictus* and *D. variabilis* larvae to determine the sensitivity of this testing method.

It is hoped that the results will be of value in future research in screening chemicals as tick repellents or attractants.

LITERATURE REVIEW

The behavioral responses of arthropods are greatly influenced by several factors such as temperature, relative humidity, light intensity, gravity, and centrifugal force. These reactions should be considered when evaluating the responses obtained from chemicals, since there may be an interaction among the mentioned factors and the chemicals. The review presented in this paper will be limited to material involving related behavioral responses to centrifugal force and in conjunction with chemical responses.

Granett *et al*. (1947) tested *Amblyonna americanum* nymphs and demonstrated that when ticks were placed on an equally illuminated circular filter paper they became uniformly distributed over the surface. They observed that the ticks exhibited an attraction toward light. The outer area of filter paper was treated, leaving the center portion free of chemicals. Ticks were placed in the center portion and counts were made on the number of ticks crossing into the treated surface. "When the treatments on both halves of the test surface were the same, each foot-candle difference in light up to 30 resulted in approximately a 2% attractiveness toward the side of the greater light intensity." He further stated, "Repellency of various chemicals to ticks under laboratory conditions can be ascertained by applying the chemical under test to a filter paper test surface."

Smith *et al*. (1946) tested *A*. *americanum* by counting the number of ticks crossing a band of skin or clothing. They concluded that the band method was inconsistent and, therefore, unsatisfactory. Lees (1948) indicated that the most characteristic reaction in orientation was a positive or negative taxis in which the ticks oriented toward or away from the stimulus.

In earlier work on behavioral response to turntable, Lyon (1900) reported that Krieal (Sitz.-Ber. der kais. Akad. der Wissench., Wien, 1893, cii, Abth. 3, p. 149.) found that a crustacean, Palceomon, reacted upon the turntable exactly like some vertebrates, running in circles opposite to the direction of rotation. Lyon (1900) also reported that Dr. Loeb (Der Heliotropismus der Thiere, etc; Anhang, Einige weitere Versuche über den Geotropismus der insekten Würzburg, Georg Hertz, 1890.) rotated houseflies, beetles, locusts, grasshoppers, dragonflies, and ants on a horizontal turntable and that they behaved exactly like vertebrates, running in circles opposite to the motion of the turntable. He stated, "while all the species tried showed compensatory motions of one or more forms, these motions ceased entirely when the eyes were thrown out of function."

Fraenkel et al. (1961) reports that Radl (1903, Leipzig. pp. 188.) observed compensatory movements in insects when placed on a turntable. The insects either rotated on one point to counteract the rotation of the table or walked or flew along a curved path. Fraenkel indicated that the above is the general effect to keep the optical field relatively constant.

Carthy (1962) reported that "in the laboratory, Winkles, Littorina neritoides, can be shown to be negatively geotactic, for in a horizontal circular dish rotated about its centre, they moved against the centrifugal force towards the axis of rotation." He further reported that razor shells, Ensis ensis and E. siliqua, will burrow downward when placed on the surface of wet sand; however, when placed in a rotating box filled with wet sand they dug in a direction which is the resultant between the centrifugal force and gravity. Their position of orientation was slightly outward and downward.

Miesch (1964) and Sterling (1966) used a turntable rotating 1 RPM in tests with cockroaches. No directional responses attributable to the turntable were reported.

Hoffman (1965) used a turntable rotating 1 1/4 RPM in membrane tests using stable flies, Stomoxys calcitrans (L). Directional responses were not reported when the flies were placed on the turntable.

Garrett (1965) used a turntable, 36 inches in diameter, rotating 1 RPM in behavioral studies using houseflies. In tracking patterns and feeding observations, he demonstrated that flies did not approach the feeding medium in any particular set pattern; however, he indicated that the flies demonstrated some restless actions characterized by circular and angular movements. He stated, "after exploring the medium with the labellum they would either stay and feed to satiation or leave the medium, make an 180^{o} turn, and return to feed. They were observed to turn at sharp angles or in circles in either direction after leaving the medium before returning to feed." Some of the flies when approaching on foot would crawl over the medium a few millimeters but would reverse their direction and return to feed.

METHOD AND MATERIALS

TEST ANIMALS

Larvae of two species of ticks were used in obtaining their responses when placed on a turntable and also in conjunction with repellents. *Dermacentor variabilis* (*Say*) replete females were collected in the fall in east central Oklahoma from hunting dogs. Other replete females were obtained in the spring from the USDA laboratory colony, Kerrville, Texas. Larvae were reared in the laboratory. *Dermacentor albipictus* replete females which had dropped to the ground in cattle holding pens were collected in late fall from Stillwater, Oklahoma. Larvae from these adults were reared in the laboratory.

The *Amblyomma maculatum* and *D. variabilis* nymphs were obtained from USDA laboratory, Kerrville, Texas. The nymphs were used only in obtaining their responses when placed on a turntable.

HOLDING CAGES

All replete females were placed three to a small pill box and stored in wide-mouthed gallon jars containing a supersaturated solution of water and ammonium nitrate to maintain a constant relative humidity. Species were kept in separate jars to facilitate testing. The lids were kept securely closed except when ticks were removed for testing.

The females were allowed to lay eggs, and the hatched larvae remained in the pill boxes until they were used in the tests.

TURNTABLE

A motor mount, constructed of 2 x 4-inch lumber, and a piece of 1-8-inch masonite, 42 inches in diameter, were fabricated into a turntable. The masonite was placed on a base plate mounted directly to the motor shaft. The 120-volt, reversible motor rotated at 2 RPM.

TEST UNIT

The test unit consisted of a 100 mm disposable plastic petri dish and filter paper. W. H. Curtin and Company, Number 7775, semi crape 90 mm, filter paper was placed into the inverted top of the petri dish. The bottom was cut out of a petri dish to form a plastic ring that would fit snugly into the inverted top of the petri dish on top of the filter paper. The ring was weighted down by clamping small strips of lead around the rim of the ring. The filter paper was divided into halves with a median line. The inner surface of the ring was coated with petroleum jelly to prevent the escape of ticks.

PEET-GRADY CHAMBER

All experiments were conducted inside a 6 x 6 x 6-ft Peet-Grady chamber modified with a plastic top to admit light. A small hot plate and a beaker of water were used to maintain the relative humidity at a range of 70 to 80%. The temperature range was between 75 and 78 F. Two rows of fluorescent lights were positioned almost directly overhead.

LABORATORY PROCEDURE

Each replicate consisted of four differently oriented test units. The turntable was divided into eight locations which were at radial intervals of 45^{o}. The center of each testing unit was placed 18 inches from the center of the turntable. The testing units were arranged in two different positions. Two units were arranged with the dividing median line of the filter paper parallel to the tangent to the circumference of the turntable (i.e., perpendicular to a radial line on the turntable) and the remaining two with the median line perpendicular to the tangent (i.e., parallel to a radial line on the turntable). The two positions were further divided. The parallel testing units were differentiated into the inward half and the outward half when oriented with the edge of the turntable. The perpendicular testing units were differentiated into the forward half and backward half when considering the direction of rotation. The ticks were transferred to a vial with a cap and then 10 were tapped out of the vial onto the predetermined half of the marked filter paper. The ticks were placed in the testing units and then set on the turntables. At the end of 15 minutes the counts were made except in a preliminary uniformity test when observations were taken at 15, 30, and 45 minutes.

All responses were measured by counting the number of ticks which were not in the semi-circle where they had been placed.

CHEMICAL MATERIALS

Five repellents diluted with acetone to make three concentrations, 0.005, 0.05, and 0.5%, were used. The following are the repellents used and their technical formulations.

1. Army Issue M-1960

N-butylacetanilide	30.00%
2-butyl-2-ethyl-1,3-propanediol. .	30.00%
Benzyl benzoate	30.00%
Emulsifier	10.00%

2. Benzyl Benzoate 100.00%

3. MGK-264

   N-(2-ethylhexyl)-bicyclo (2.2.1)-
   hept-5-ene-2,2-dicarboximide 100.00%

4. R-11

   2,3,4,5-bis(-2-butylene)
   tetrahydrofurfural 100.00%

5. Army Issue M-6

   N,N-diethyl-m-toluamide 75.00%
   Ethanol 25.00%

PRELIMINARY TRIALS

Four parallel and four perpendicular units as described earlier were equally spaced on the turntable. In the first preliminary trial *D. variabilis* larvae were placed on the outward half of the parallel units and on the backward half of the perpendicular units. Rotation of the turntable was clockwise. The filter papers were not treated with any chemical. This test was run 10 times without randomization.

PREFERRED ORIENTATION WITHIN THE PARALLEL AND PERPENDICULAR UNITS

The two parallel units were oriented in such a way that the *D. variabilis* larvae were placed on the outward half and counts were made of the ticks observed in the inward half of the unit and on the remaining unit the ticks were placed on the inward half and counts were made of the ticks observed in the outward half of the unit. The two perpendicular units were oriented in such a way that ticks were placed on the forward half and counts made of those found in the backward half. In the remaining units ticks were placed on the backward half and counts were made on the opposite half. The rotation of the turntable was clockwise. The filter papers were not treated with any chemical. Duplicate units run at the same time were conducted for each of the replicates. The units were placed on the turntable in a completely randomized design. Counts were made at time intervals of 15, 30, and 45 minutes.

ORIENTATION TO CLOCKWISE VERSUS COUNTER CLOCKWISE ROTATION

This test was identical to the test immediately above except that the rotation of the turntable was reversed.

ABSENCE OR PRESENCE OF "TICK FACTOR"

Four testing units with *D. variabilis* larvae were restricted to half of the units overnight. The halves containing the ticks overnight were marked and counts were made on the marked half only. At the beginning of each replicate ticks were placed on the opposite side of the marked half. In four other units, unused filter paper was placed in each unit before each replicate. The parallel and perpendicular positions were used as described in the above tests. The rotation of the turntable was counter clockwise. Duplicate units run at the same time were used for each of the 10 replicates.

FURTHER TESTING WITH *D. ALBIPICTUS* AND *D. VARIABILIS* LARVAE

Another preliminary test was made using two species. Separate tests were conducted on each of the two species. Each test consisted of 10 replicates, each with duplicate samples. The two factors being investigated were the position of the testing units (parallel and perpendicular) and direction of orientation within the two positions of the test units.

NYMPHAL ORIENTATION TEST

This test was conducted similarly to tests using larval ticks.

Rotation of the turntable was counter clockwise. Duplicate tests run at the same time were used for each of the 10 replicates. The nymphs tested were D. variabilis and A. maculatum. The tests were conducted separately for each species. In the test using A. maculatum, only 120 individuals were available for testing, and these nymphs were used for the 10 replicates. A new randomization of the units was used for each duplicate. D. variabilis nymphs were used once in each replicate and were discarded. Location of the position of the units on the turntable were randomized for each replicate.

LARVAL RESPONSE TO REPELLENTS

This test was run five times using five repellents each at three concentrations, 0.005, 0.05, and 0.5%. Two species, D. variabilis and D. albipictus, were used simultaneously on the turntable. The two parallel and two perpendicular positions were used for each species, giving 8 units on the turntable. Filter papers for each replicate were dipped in the chemical solution to the median line and allowed to dry for one hour before testing. The procedures used in testing each chemical were as follows: (1)the first test was with filter papers treated with acetone only; (2) the second test was with filter papers treated with 0.005% of the test chemical; (3) the third test was with filter papers treated with 0.05%; and (4) the fourth test was with filter papers treated with 0.5%. The five repellents were tested in a random order for each block. The orientation of the units was randomized to occupy the eight equally spaced locations on the turntable numbered 1 through 8. This test was a randomized complete block experiment, having a factorial arrangement of 2 species, 5 repellents, 3 levels of concentration, 2 levels of positions (parallel and perpendicular) and two levels of direction of orientation.

RESULTS AND DISCUSSION

PRELIMINARY TRIALS

The analysis of variance indicated that there were highly significant differences in the responses between the parallel and perpendicular units. An average of 3.45 of the 10 ticks placed on the outward half of the parallel unit moved to the inward half of the parallel unit. An average of 5.52 of the ticks placed on the backward half of the perpendicular unit moved to the forward half.

The results of this test indicated a need for further testing to determine the orientation responses of ticks.

PREFERRED ORIENTATION WITHIN THE PARALLEL AND PERPENDICULAR UNITS

No significant differences were noted between the parallel and perpendicular positions; however, there was a highly significant difference in the direction the ticks oriented themselves within the two positions. The orientation for the parallel unit was significantly outward toward the edge of the turntable. Ticks placed on the inward half migrated in far larger numbers toward the outer half than those placed in the outer half migrating toward the inward half. The orientation for the perpendicular units was more in the direction of rotation (Table I). This test indicated that this species of tick oriented outward toward the edge of the turntable and slightly forward in the direction of rotation (Figure 1a).

There also was a highly significant difference between positions x

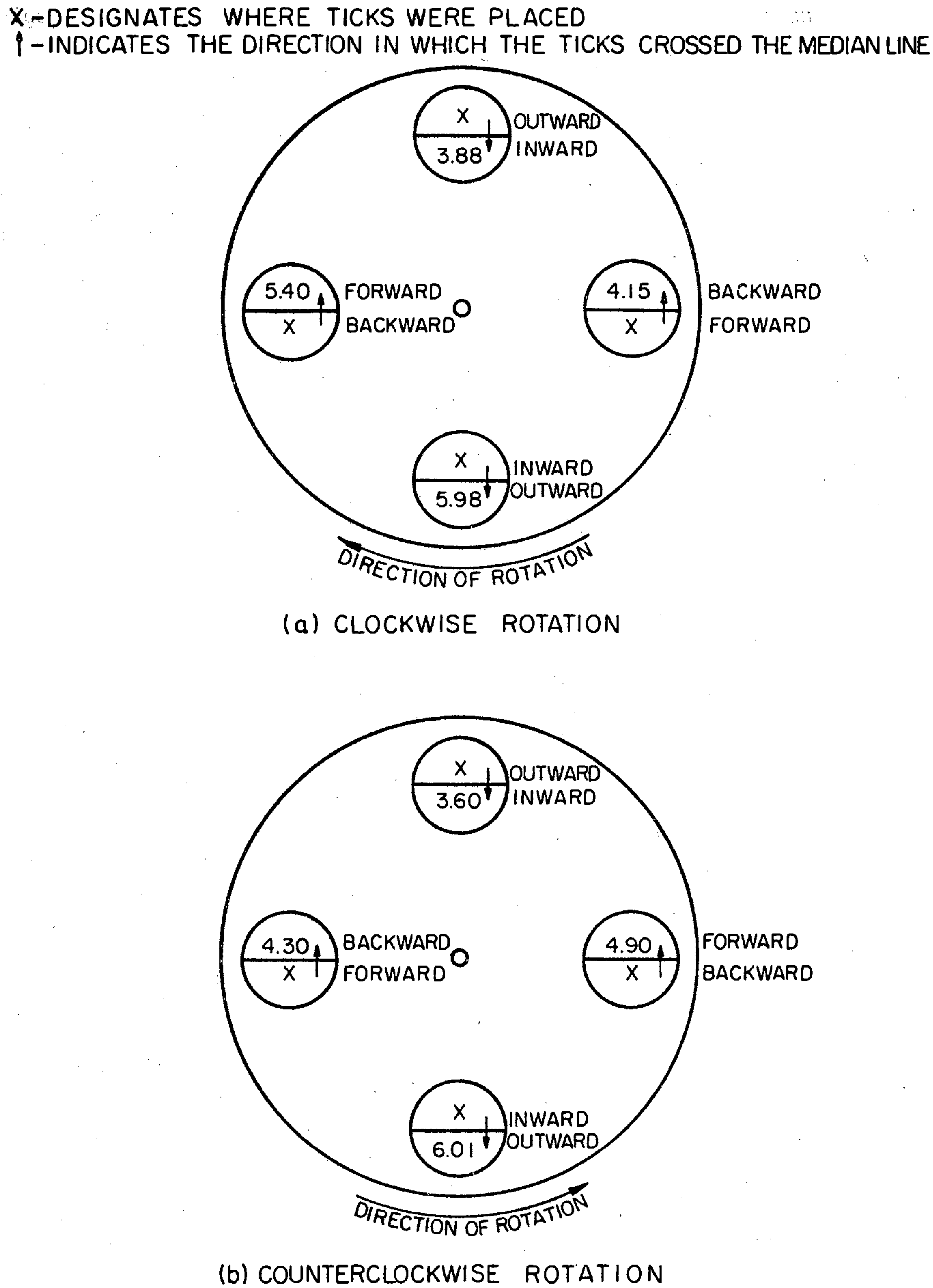


Figure 1. Directional responses due to the effects of rotation expressed in mean numbers of ticks that were across the median line at the time of counting.

the directions of orientation. This indicates an interaction between the above factors. When the ticks move from the outward half to the inward half, there is less directional response than when the ticks move from the forward half to the backward half and vice versa.

A time difference test was run in conjunction with this experiment. The AOV indicated no significant difference between the time intervals of 15, 30, and 45 minutes for making counts. This test established that all readings could be taken 15 minutes after the start of each test.

ORIENTATION TO CLOCKWISE VERSUS COUNTER CLOCKWISE ROTATION

This test was conducted to determine if measurable differences in responses could be detected if the rotation of the turntable was reversed. The results of the AOV were almost identical to the AOV of the clockwise rotation, indicating a highly significant difference in the direction of orientation of the tick and also indicating a similar interaction between positions x direction of orientation (Table I).

TABLE I

DIRECTIONAL RESPONSES EXPRESSED IN MEAN NUMBERS OF TICKS THAT WERE ACROSS THE MEDIAN LINE IN THE CLOCKWISE AND COUNTER CLOCKWISE ROTATION TESTS

	Position of Units			
	Parallel		Perpendicular	
Rotation	Outward	Inward	Forward	Backward
Clockwise	5.95	3.88	5.40	4.15
Counter clockwise	6.01	3.60	4.90	4.30

As can be seen in Figure 1b, the ticks only changed direction of orientation within the perpendicular units and that was in the direction of rotation. A time difference factor was included in this test and the results were similar to those in the clockwise rotation test.

ABSENCE OR PRESENCE OF "TICK FACTOR"

This test was initiated to determine possible "factors" remaining after ticks had been removed from the filter paper where they had been for 16 hours. The results of this test indicated no significant difference between the filter papers.

NYMPHAL ORIENTATION TEST

Using the same test procedures as used in larval tests, difficulties were encountered in keeping a few ticks within the test unit. They oriented outward toward the edge of the turntable and forward in the direction of rotation (Table II) as shown in larval test.

TABLE II

DIRECTIONAL RESPONSES EXPRESSED IN MEAN NUMBERS OF TICKS THAT WERE ACROSS THE MEDIAN LINE WHEN NYMPHS, *A. MACULATUM* AND *D. VARIABILIS*, WERE TESTED

	Position of Units			
	Parallel		Perpendicular	
Species	Outward	Inward	Forward	Backward
A. maculatum	4.95	3.55	4.60	4.25
D. variabilis	5.30	4.05	4.60	4.05

FURTHER TESTING WITH *D. ALBIPICTUS* AND *D. VARIABILIS*

Tests were conducted separately for each species and the results were similar. Both AOV indicated a highly significant difference in direction of orientation within the two positions. No significant difference was indicated for positions, but positions x direction of orientation were highly significant (Table III).

TABLE III

DIRECTIONAL RESPONSES EXPRESSED IN MEAN NUMBERS OF TICKS THAT WERE ACROSS THE MEDIAN LINE WHICH SHOWS THE SIMILAR INTERACTION FOR EACH SPECIES

	Position of Units			
	Parallel		Perpendicular	
Species	Outward	Inward	Forward	Backward
D. albipictus	5.00	2.85	4.35	3.10
D. variabilis	6.00	3.30	4.90	4.55

LARVAL RESPONSE TO REPELLENTS

The results of the test were broken down into a split plot AOV (Table XII). The main plots were treatment combinations which included controls, concentrations, and repellents, whereas the subplots were species, position, and direction of orientation. Within the main plots the following were highly significant: (1) controls versus chemical treatments; (2) repellents; (3) concentrations. The difference in repellents x concentrations were significant.

Within the subplots the highly significant factors were: (1) Direction of orientation; (2) direction of orientation x (controls versus chemical treatments); (3) direction of orientation x concentrations; (4) species x direction of orientation; (5) species x direction of orientation x (controls versus chemical treatments);(6) positions x direction of orientation; (7) position x direction of orientation x concentrations; and (8) position x species x direction of orientation. Those factors which were significant (P = 0.5) were: (1) positions x direction of orientation x (controls versus chemical treatments); (2) position x direction of orientation x controls; and (3) position x species x direction of orientation x concentration.

The difference in responses to the five repellents were demonstrated by the table of means (Table IV).

TABLE IV

THE AVERAGE NUMBER OF TICKS THAT WERE ACROSS THE MEDIAN LINE ON THE CHEMICALLY TREATED FILTER PAPERS FOR EACH REPELLENT

Repellent	Mean Numbers
R-11	1.77
N,N-diethyl-m-toluamide	2.24
Benzyl benzoate	2.39
M-1960	2.39
MGK-264	2.75

The results of this test indicated that R-11 is more repellent to ticks than the remaining four. Steelman (1963) tested R-11 and MGK-264 in filter paper tests and indicated that the former gave greater repellency, which is also indicated in this test. In a similar test he showed M-1960 to have a greater repellency value against _A. americanum_ larvae than N,N-diethyl-m-toluamide. However, using the test method involving a turntable, the results indicated that N,N-diethyl-m-toluamide is slightly more repellent than M-1960 when tested at equal concentrations. Taylor (1960) using the patch test indicated that R-11 at concentrations of 0.1, 0.25, and 1.0% gave 100% repellency after the first day of treatment against Rhipicephalus sanguineus, whereas MGK-264 at the same concentrations gave 99, 86, and 93% repellency. Gouck et al. (1955) demonstrated that M-1960 gave an average of 95% repellency for one to three weeks as a clothing impregnation against A. americanum, whereas crude N,N-diethyl-m-toluamide gave an average of 90%.

As expected, a highly significant difference was noted between the concentrations. An interaction was indicated by the significance of repellents x concentrations (Table V).

TABLE V

THE INTERACTION OF REPELLENTS X CONCENTRATIONS IS SHOWN BY THE AVERAGE NUMBER OF TICKS THAT WERE ACROSS THE MEDIAN LINE FOR EACH OF THE REPELLENTS AND THEIR CONCENTRATIONS

	Repellents				
Concentrations	R-11	DET[a]	B.B.[b]	M-1960	MGK-264
0.005	3.67	4.62	5.02	4.62	5.35
0.05	1.47	1.87	2.07	2.42	2.92
0.5	0.17	0.22	0.07	0.12	0.00

a = N,N-diethyl-m-toluamide

b = Benzyl benzoate

The responses of the two species demonstrated no significant differences between their reactions to repellents, concentrations, and the combination of repellents x concentrations. The two species showed no significant difference among the controls which really constitute a uniformity trial.

The highly significant difference in direction of orientation x concentrations indicated an interaction between the two factors (Table VI).

TABLE VI

THE INTERACTION OF DIRECTION OF ORIENTATION X CONCENTRATIONS IS SHOWN BY THE AVERAGE NUMBER OF TICKS THAT WERE ACROSS THE MEDIAN LINE FOR EACH CONCENTRATION AND DIRECTION OF ORIENTATION

	Direction of Orientation	
Concentrations	Forward & Outward	Backward & Inward
0.005	5.17	4.15
0.05	2.53	1.78
0.5	0.11	0.13

For factors, species x direction of orientation, there was a highly significant difference which indicated an interaction (Table VII).

TABLE VII

THE INTERACTION OF SPECIES X DIRECTION OF ORIENTATION IS SHOWN BY THE AVERAGE NUMBER OF TICKS THAT WERE ACROSS THE MEDIAN LINE FOR EACH SPECIES AND DIRECTION OF ORIENTATION

	Direction of Orientation	
Species	Outward & Forward	Inward & Backward
D. variabilis	3.25	2.98
D. albipictus	3.59	2.28

Direction of orientation was highly significant and followed the uniformity trial patterns. When the ticks were subjected to repellents as well as no repellents, the orientation of the ticks was toward the outside edge going with the centrifugal force, and slightly forward in the direction of rotation of the turntable (Table VIII).

TABLE VIII

DIRECTIONAL RESPONSES EXPRESSED IN MEAN NUMBERS OF TICKS THAT WERE ACROSS THE MEDIAN LINES FOR ORIENTATIONS OF (1) OUTWARD AND FORWARD AND (2) BACKWARD AND INWARD FOR AMONG CONTROLS, REPELLENTS WITH CONTROLS, AND REPELLENTS

Direction of Orientation	Among Controls	Repellents With Controls	Repellents
Outward and Forward	5.77	3.39	2.60
Backward and Inward	4.46	2.63	2.02

Position of units x direction of orientation was highly significant and indicated an interaction between the two factors (Table IX).

TABLE IX

THE INTERACTION OF POSITION OF UNITS X DIRECTION OF ORIENTATION IS SHOWN BY THE AVERAGE NUMBER OF TICKS THAT WERE ACROSS THE MEDIAN LINE FOR EACH POSITION OF UNITS AND DIRECTION OF ORIENTATION

	Direction of Orientation	
Position of Units	Outward & Forward	Backward & Inward
Parallel	3.62	2.40
Perpendicular	3.17	2.85

The significance of position of units x direction of orientation x concentrations indicated an interaction between the three factors. From the lower concentration to the higher there was a difference in response for the position x direction of orientation (Table X).

TABLE X

THE INTERACTION OF POSITION OF UNITS X DIRECTION OF ORIENTATION X CONCENTRATIONS IS SHOWN BY THE AVERAGE NUMBER OF TICKS THAT WERE ACROSS THE MEDIAN LINE FOR EACH OF THE ABOVE FACTORS

	Direction of Orientation			
	Parallel		Perpendicular	
Concentrations	Outward	Inward	Forward	Backward
0.005	5.50	3.66	4.84	4.64
0.05	2.70	1.72	2.36	1.84
0.5	0.12	0.18	0.10	0.08

The interaction of factors, position of units x species x direction of orientation, indicated that the responses of the two species were different when position and orientation of units were considered (Table XI).

D. albipictus larvae which are usually in a quiescent stage by late May exhibited a greater response in moving from the inward half to the outward half of the parallel units than did D. variabilis larvae.

The filter papers were dipped so that the paper was wetted to the median line. Sometimes the solution crept slightly past the median line, but counts were made only of those ticks that were across the median line at the end of 15 minutes.

TABLE XI

THE INTERACTION OF POSITION OF UNITS X SPECIES X DIRECTION OF ORIENTATION IS SHOWN BY THE AVERAGE NUMBER OF TICKS THAT WERE ACROSS THE MEDIAN LINE FOR EACH OF THE ABOVE FACTORS

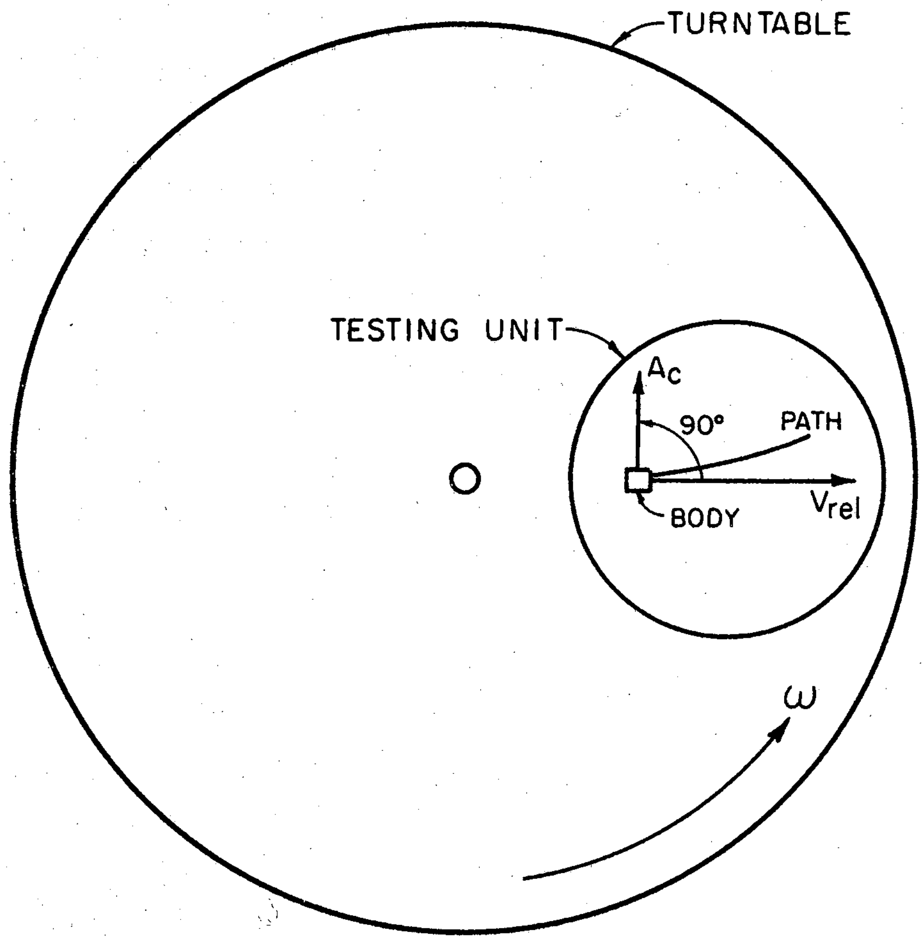
	Direction of Orientation			
	Parallel		Perpendicular	
Species	Outward	Inward	Forward	Backward
D. variabilis	3.08	2.95	3.32	3.01
D. albipictus	4.16	1.86	3.02	2.70

At higher concentrations ticks were observed to orient themselves along the border of the treated portion without crossing onto the treated surfaces. It was also noted that a tick which got on the treated surfaces containing high concentrations of the repellent became desensitized to the repellent and wandered on and off the treated surface without regard to the border.

PROBABLE CAUSES OF ORIENTATION ON A TURNTABLE

Since the ticks prefer to orient significantly outward with the centrifugal force and slightly forward in the direction of rotation, regardless of clockwise or counter clockwise rotation, it is hypothesized that the orientation in the direction of rotation may be due to the ticks' responding to coriolis acceleration.

Coriolis acceleration will result when a body moves on a path which lies on a rotating platform. The direction of coriolis acceleration on the body (tick) is found by rotating the relative velocity direction (Vrel) 90° in the direction of the turntable velocity (ω). This is shown in Figure 2. If the body is moving on a path which is a straight



ω = ROTATION OF TURNTABLE

V_{rel} = VELOCITY OF BODY RELATIVE TO THE TURNTABLE

A_c = CORIOLIS ACCELERATION

Figure 2. Illustration of coriolis acceleration

line, the resulting coriolis acceleration would tend to cause the body to deviate from its straight-line motion, as discussed above.

If the orientation in the direction of rotation is not a response due to coriolis acceleration, an alternate explanation may be that they are responding to the movement of air caused by their movement through a body of air at a constant velocity.

SUMMARY AND CONCLUSIONS

The behavioral responses of *D. albipictus* and *D. variabilis* larvae and *A. maculatum* and *D. variabilis* nymph ticks placed on a turntable rotating at 2 RPM were obtained. Uniformity trial tests were conducted separately for each species and developmental stages. The uniformity trials indicated a highly significant preference of the ticks to orient outward toward the edge of the turntable and slightly forward in the direction of rotation.

All tests were set up in a factorial arrangement of treatments. In the uniformity trials the factors being investigated were time the ticks were on the test unit, position of test units (parallel or perpendicular orientation of the median line to the tangent), and direction of orientation within the position of test units. In the other tests the factors investigated were repellents, concentrations, species, position of test units, and direction of orientation. The behavioral responses in the latter test were similar to those obtained in the uniformity test but of less magnitude.

A difference was shown among the repellents tested: R-11, N,N-diethyl-m-toluamide, benzyl benzoate, M-1960 and MGK-264. R-11 was the most effective chemical in repelling larval ticks, followed by N,N-diethyl-m-toluamide.

At higher concentrations ticks were observed to orient along the border of the treated area, seldom getting on the treated surface; however, when a tick did get on a treated surface it became desensitized to the repellent.

The interaction of repellents x concentration indicated that the responses for each repellent were not the same from the higher concentration to the lower concentration.

The results of these tests demonstrated the effectiveness of the turntable as a tool in evaluating repellents or attractants when the behavioral responses of larval ticks were considered.

It is felt that the use of all four test unit orientations gave a better measurement of responses to the materials being evaluated.

The orientation forward in the direction of rotation may be due to coriolis acceleration.

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APPENDIX

TABLE XII

ANALYSIS OF VARIANCE OF TICK MOVEMENT DUE TO MOTION AND CHEMICAL TREATMENT

Source	D. F.	M. S.	F (calc)	$F_{.05}$	$F_{.01}$
Total	799				
Main Plots	99				
Replicates	4	2.9031			
Treatment Combinations and Controls					
Control vs Chemical Treatments	1	1,178.8016	397.04	--	6.99
Among Controls	4	0.5700	0.19	2.49	--
Among Treat. Comb.					
Repellents	4	15.1566	5.10	--	3.58
Concentrations	2	1,034.2616	348.36	--	4.89
Repellents x Concentrations	8	6.5491	2.21	2.06	2.75
Error (a)	76	2.9689			
Subplots	700				
Species	1	4.8050	2.44	3.84	--
Species x Treatment Combinations and Control					
Species x (Control vs Chem. Treat.)	1	3.3750	1.71	3.84	--
Species x Control	4	1.7700	0.90	2.37	--
Species x Chem. Treat.					
Species x Repellents	4	1.2433	0.63	2.37	--
Species x Concentrations	2	2.7150	1.38	3.00	--
Species x Repellents x Conc.	8	3.1983	1.62	1.94	--
Position	1	0.0000	0.00	3.84	--

TABLE XII, Continued

Source	D. F.	M.S.	F (calc)	$F_{.05}$	$F_{.01}$
Position x Treat. Comb. and Control					
Position x (Cont. vs Chem. Treat.)	1	0.0066	0.01	3.84	--
Position x Controls	4	1.4300	0.73	2.37	--
Position x Chem. Treat.					
Position x Repellents	4	0.8100	0.41	2.37	--
Position x Concentrations	2	1.0316	0.52	3.00	--
Position x Repellents x Conc.	8	0.9400	0.47	1.94	--
Favor {(outward + forward) vs (inward + backward)}	1	117.0450	59.39	--	6.63
Favor x Treat. Comb. and Control					
Favor x (Cont. vs Chem. Treat.)	1	22.6950	11.51	--	6.63
Favor x Controls	4	4.5300	2.29	2.37	--
Favor x Chem. Treat.					
Favor x Repellents	4	0.8100	0.41	2.37	--
Favor x Concentrations	2	14.5616	7.38	--	4.61
Favor x Repellents x Concentrations	8	1.0533	0.53	1.94	--
Species x Position	1	4.5000	2.28	3.84	--
Species x Position x Treat. Comb. and Controls					
Species x Position x (Cont. vs Chem. Treat.)	1	0.9600	0.48	3.84	--
Species x Position x Control	4	1.1200	0.56	2.37	--
Species x Position x Chem. Treat.					
Species x Position x Repellents	4	2.2566	1.14	2.37	--
Species x Position x Conc.	2	1.0850	0.55	3.00	--
Species x Position x Repellents x Conc.	8	2.0391	1.03	1.94	--
Species x Favor	1	59.4050	30.14	--	6.63

TABLE XII, Continued

Source	D. F.	M. S.	F (calc)	$F_{.05}$	$F_{.01}$
Species x Favor x Treat. Comb. and Controls					
Species x Favor x (Cont. vs Chem. Treat.)	1	18.3750	9.32	--	6.63
Species x Favor x Control	4	4.0700	2.06	2.37	--
Species x Favor x Chem. Treat.					
Species x Favor x Repellents	4	0.4433	0.22	2.37	--
Species x Favor x Conc.	2	5.2550	2.66	3.00	--
Species x Favor x Repellents x Conc.	8	2.8008	1.42	1.94	--
Position x Favor	1	17.0016	8.62	--	6.63
Position x Favor x Treat. Comb. and Controls					
Positions x Favor x (Cont. vs Chem. Treat.)	1	7.7066	3.91	3.84	6.63
Position x Favor x Controls	4	5.0300	2.55	2.37	3.32
Position x Favor x Treat. Comb.					
Position x Favor x Repellents	4	1.0266	0.52	2.37	--
Position x Favor x Concentrations	2	9.6717	4.90	--	4.61
Position x Favor x Repellents x Conc.	8	1.4091	0.71	1.94	--
Position x Species x Favor	1	58.3200	29.59	--	6.63
Position x Species x Favor x Treat. Comb. and Controls					
Position x Species x Favor x (Cont. vs Chem. Treat.)	1	5.6066	2.84	3.84	--
Position x Species x Favor x Controls	4	2.1200	1.07	2.37	--
Position x Species x Favor x Chem. Treat.					
Position x Species x Favor x Repellents	4	0.3566	0.18	2.37	--
Position x Species x Favor x Conc.	2	7.4316	3.77	3.00	4.61
Position x Species x Favor x Repellents x Conc.	8	1.0066	0.51	1.94	--
Error (b)	560	1.9706			

VITA

Jimmy L. Young

Candidate for the Degree of

Master of Science

Thesis: BEHAVIORAL RESPONSES OF TICKS TO MOTION AND REPELLENTS OR ATTRACTANTS

Major Field: Entomology

Biographical:

Personal Data: Born in Cement, Oklahoma, April 1, 1938, the son of James H. and Orpha A. Young.

Education: Undergraduate study at Cameron State Agricultural College, Lawton, Oklahoma, 1957-59; Bachelor of Science degree in Entomology from Oklahoma State University, Stillwater, Oklahoma, 1961. Graduate study, Oklahoma State University, 1963, 1967-68.

Professional Experience: Farming and animal husbandry previous to 1957; Entomology Department, Oklahoma State University, 1960-61; Medical Entomologist, Medical Service Corps, United States Army, 1961-62; Tulsa City-County Health Department, 1964; 3rd. U. S. Army Medical Laboratory, 1964; Medical Entomologist, Medical Service Corps, U. S. Army, 1964 to present.

Member of the Sigma Xi, Sanborn Entomology Club.